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CONTRACTING ORGANIZATION: Henry M. Jackson Foundation for the Advancement
of Military Medicine
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14. ABSTRACT

Objective: To increase our understanding of the molecular aberrations associated with endometrial carcinogenesis and the biologic mechanisms underlying the protective effect of oral contraceptive (OC) therapy. **Methods:** 1) Oligonucleotide microarray analysis was performed on a panel of endometrial cancers. 2) A subset of adenocarcinoma cases from the International DES Registry (IDESR) was analyzed for MSI 3) A case-control study of the CASH database was performed to evaluate the relationship between progestin potency and endometrial cancer risk. 4) An analysis of endometrium samples from cymologous macaques that were exposed to long term progestins was performed. 5) A clinical trial comparing progestin versus placebo is underway that will facilitate investigation of the effects of progestin exposure on the endometrial lining. **Results:** 1) Different histological types of endometrial cancer have unique genomic expression patterns. 2) The poor quality DNA from the majority of IDESR samples prohibited an adequate analysis of the case set. 3) A case-control study has suggested higher progestin- potency OCs may be more protective than lower progestin potency OCs among women with a larger body habitus. 4) Macaque studies have suggested that induction of apoptosis may be a mechanism underlying the chemoprotective effects of progestin on the endometrium. 5) Regulatory hurdles have resulted in delays in initiation of the clinical trial. Final FDA approval is expected within the next 3-4 months and the original objectives in the statement of work will be addressed.

15. SUBJECT TERMS
Endometrial cancer, hormone, prevention

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INTRODUCTION

Endometrial cancer is the most common type of gynecologic cancer in the United States and was estimated by the American Cancer Society to have been newly diagnosed among 40,000 American women in the year 2004 and lead to approximately 6500 cancer related deaths (1). Approximately 25% of all endometrial cancers occur in premenopausal women (2). Major advances in our understanding and treatment of endometrial cancer have occurred over the past decade, yet the frequency of this cancer in the general population has not been altered appreciably. Despite the known protective effect of oral contraceptives, little has been learned regarding the underlying mechanism. We believe that an understanding of the molecular profiles of endometrial cancers and the molecular events underlying the protective effect of oral contraceptives against endometrial cancer could facilitate the development of effective chemopreventives and significantly decrease the incidence of endometrial cancer in women.

BODY

Aim 1: To characterize and compare the molecular profiles of Type I endometrioid endometrial cancers, which often develop in an estrogen milieu, to that of Type II endometrial cancers. In addition, we will use microarray to examine the molecular changes in the endometrium associated with progestin exposure in order to gain insight into the biologic mechanism underlying the chemopreventive effect of the oral contraceptive pill (OCP).
Project 1: Objectives completed and data previously submitted with 2004 report. Data published this past year and listed in "Reportable Outcomes".

Project 2 (Pending): The short-term effects of progestins gene expression in the endometrium will be evaluated using uterine specimens collected from patients enrolled in a double-blinded prospective randomized trial. This study was first submitted to the Department of Clinical Investigation (DCI), which functions as the local IRB for all studies being conducted at Walter Reed Army Medical Center (WRAMC), on 11/26/01. DCI reviewed the protocol and primarily approved it on 1/11/02. Approval by the Clinical Investigation Regulatory Office for the Dept of Army was received 2/21/02. As this study is being funded under the Molecular Biology and Prevention of Endometrial Cancer grant, the protocol was then transferred to the Office of Research Protections (ORP), US Army Medical Research and Material Command, for second level review of primary protocols at both WRAMC and Evanston Northwestern (EN). The protocol was also submitted to the IRB at Evanston Northwestern for review/approval and all documents were then transferred to ORP for second level review and approval. Delays in approval by both the primary and second level review authorities resulted in having the two separate IRB groups agreeing on certain points of the protocol where there was initial disagreement. Approval from ORP was finally received on 3/21/05. Approval from both the local and second level IRB was required before clinical work under the protocol was allowed to commence. By the time both protocols received local and second level IRB approval approximately three years later on March 21, 2005, Wyeth had ceased distribution of Ovrette in the US. The decision to cease distribution of Ovrette in the U.S. was purely a marketing decision and is not reflective of any safety-related issues associated with the drug or the manufacturing of the drug.

Wyeth was contacted on March 29, 2005 regarding the ability to gain access to the drug for use in our clinical trial. Staff worked with Mary Dorr, Senior Director, Wyeth Global Medical Affairs, to investigate the feasibility of obtaining the drug from Canada at no charge. After further investigation,

we discovered that the drug is currently manufactured in Canada solely for export to underdeveloped countries under a government aid program. We are not able to import this drug into the U.S. for our clinical trial for a number of reasons, including the fact that the manufacturing plant in Canada has not received approval from the U.S. Food and Drug Administration (FDA) to manufacture this drug. After discovering that Ovrette was no longer available in the U.S., we contacted the FDA to inquire about possible alternatives. We were told that that we would need to find a supplier of the active ingredient who has a Drug Master File (DMF) filed with the FDA. After several months, we located a company in China that would provide levonorgestrol in bulk as well as provide the DMF to the company formulating the drug and placebo. Additionally, we located a company that will provide all analytical testing of the drug and placebo required for regulatory approval. Once a contract is signed, the drug will be formulated and tested in the ensuing one/two months. Documentation of all processes and tests will have to be compiled and submitted to the FDA in the form of an information amendment to the Chemistry Manufactures and Controls (CMC) section of an Investigational New Drug (IND) application. (An amendment to the clinical study plan would also be submitted at the same time to include this study under the IND.) The FDA has 30 days to review an IND submission and provided there are no concerns, we would be approved to commence the study.

The study will have to be submitted again to both the local and second level IRBs, documenting that this is now an investigational study being conducted under an approved IND. We hope that this process will proceed swiftly as this has been a significant source of delay in the past. It is note worthy to mention that there should not be any changes to the study protocol—unless requested by the FDA—with the exception that this is now an investigational study being conducted under an approved IND.

We have officially received a one-year "No Cost Extension," extending the performance period to 17 July 2007, with research ending 17 June 2006. If everything goes as planned, we will begin enrollment in the study in October/November 2006. The study requires the enrollment of 82 participants, 41 at each site, which we feel we should be able to achieve in an 8-12 month time frame. Research on the specimens would then have to be conducted and analyzed, which would most likely take an additional 3-6 months after enrollment.

Aim 2: To analyze vaginal and cervical adenocarcinomas, that have arisen in women exposed to DES in-utero, for methylation and mutation of PTEN and MLH1 in order to determine if estrogen induces genetic alterations in these tumors characteristic of Type I endometrioid carcinomas.
Although a pilot study aimed at an analysis of MSI in 7 cases from the International DES Registry was successful with repetitive attempts at DNA amplification, the analysis of the entire set was not successful presumably secondary to the quality of the DNA which reflects the old age of the specimens and the various methods that were used in their preservation. Less than 50% of the samples amplified at any one of the markers making the data inadequate for designation of MSI status. Significant amounts of material from the Transplacental Registry were used unsuccessfully to complete the work on the microsatellite instability.   Acquisition of additional material to further evaluate alterations in either mismatch repair genes (causative of MSI) or PTEN was not an option.

Aim 3: Using data from the Centers for Disease Control Cancer and Steroid Hormone Study, we will determine if the protective effect of OCP's against endometrial cancer are impacted by the progestin or estrogen potency of OCP formulations.

Objectives completed and data previously submitted with 2004 report. Published manuscript listed in "Reportable Outcomes"

Aim 4: To test the hypothesis that the oral contraceptives and hormone replacement therapy progestins provide a chemoprotective effect against endometrial cancer through induction of apoptosis, PTEN, and TGF-beta in the endometrium.
Epidemiological studies have demonstrated that OCP use lowers the risk of subsequent endometrial and ovarian cancer. Although the biologic mechanism(s) underlying the protective effect of OCP's on the risk of both of these cancers have not been well defined, there is evidence to suggest that biologic effects related to the progestin component may underlie the cancer preventive effects of the OCP.   Recent studies have reported the progestin-mediated activation of apoptosis in endometrial cancer cell lines and endometrial hyperplasias. The finding that progestin activates the apoptosis pathway in endometrial cells raises the possibility that this may be a major mechanism underlying the therapeutic effect of progestins against endometrial hyperplasia. Similarly, our group has found that progestins markedly activate both apoptosis and TGF-beta expression in the ovarian epithelium leading to the hypothesis that progestins may act as chemopreventives for ovarian cancer (see preliminary data below). It is interesting that tumors arising from the ovary and endometrium share common epidemiological risk factors, and that both the endometrium and ovarian surface epithelium share a common embryological precursor. It is thus plausible that progestins activate similar molecular pathways relevant to cancer prevention in both of these organ sites. Recent evidence suggests that expression *PTEN* appears to be upregulated in the secretory phase of the menstrual cycle. It is plausible that the chemopreventive effects of OCP's are mediated through overexpressed *PTEN* with resultant suppression of cell cycle progression and activation of apoptosis in endometrial cells.

- The short-term effects of progestins on apoptosis as well as the expression of *PTEN* and TGF-β in the endometrium will be evaluated using uterine specimens collected from patients enrolled in a double-blinded prospective randomized trial. See Aim1, project 2 for explanation of delays in deliverables and current "no cost extension status".
- The long term effects of progestins on apoptosis as well as the expression of PTEN and TGF-b in the endometrium were evaluated using uterine specimens from cynomolgus macaques (80 premenopausal and 130 postmenopausal) previously part of a three-year randomized trial designed to evaluate the effects of the combination oral contraceptive pill and hormone replacement therapy on reproductive organs. The staining of the macaque slides for PTEN was completed but interpretation of the set of slides that was stained in 3 separate runs was found on review of the slides to be less than optimal for interpretation, due to inconsistencies in the staining intensity of replicate samples and the equivocal staining and loss of PTEN staining in the macaque samples. Our suspicion is that the inconsistencies in staining may reflect the human PTEN antibody being used on the macaque tissue. Several samples were sent to Dr. Carl Morrison at Ohio State University for independent immuohistochemical analysis. Dr. Morrison's lab performed immunohistochemical staining for several sentinel papers involving the PTEN tumor suppressor gene and cancer. His lab confirmed limitations of the commercially available PTEN antibody in staining macaque tissues, and we subsequently abandoned this component of Aim 4, project 1.
Two groups of animals used to evaluate the effects of progestin containing hormonal formulations on apoptosis are presented separately in the results section of this annual report. Five-micron sections of endometrium were stained for expression of cleaved caspase 3 using

a mouse monoclonal antibody to cleaved caspase-3 (Cell Signalling Technology, Beverly, USA) and standard biotin-streptavidin immunohistochemical techniques. Immunostaining was graded within superficial endometrial glands, basal endometrial glands, superficial endometrial stroma, and basal endometrial stroma by an experienced observer blinded to treatment group. Staining was scored as absent (0), or mild (grade 1), moderate (grade 2) or marked (grade 3). Staining for cleaved caspase 3 was compared to staining for other markers of endometrial function such as histopathological assessments and expression of the proliferation marker Ki67 and sex steroid receptors progesterone receptor and estrogen receptor alpha. Detailed study designs and results are given below by study.

Nonhuman Primate Oral Contraceptive Study

Methods
Premenopausal, female cynomolgus monkeys, mean age 4.75 years at start.
↓

Randomization on the basis of body weight to 4 groups:

Treatment		N
Control (intact, normally cycling)	Group 1	20
Triphasil	Group 2	21
Cyclic ethinyl estradiol	Group 3	21
Cyclic levonorgestrel	Group 4	19

Drug doses for Hormone-treated animals:

Ethinyl estradiol	EE	Days 1-6: 0.03 mg
		Days 7-11: 0.04 mg
		Days 12-21: 0.03 mg
		Days 22-28 : no drug
Levonorgestrel	LN	Days 1-6: 0.05 mg
		Days 7-11: 0.075 mg
		Days 12-21: 0.125 mg
		Days 22-28: no drug

↓

Treatment was given for 35 months. Animals received a moderately atherogenic diet.
↓

Euthanasia and necropsy of groups 1-4. All necropsies were on the peak progestin week of treatment.
↓

Tissues were fixed in 4% paraformaldehyde and processed into paraffin for routine histology and immunohistochemistry.

Results

<u>Routine Histology</u>
All slides were examined by a board-certified veterinary pathologist in consultation with
MD gynecologic pathologists. Standard diagnostic terminology was used.

Histologic Features of Endometrium

Group	Normally cyclic	Atrophic	Hyperplastic	Disordered	Cancer
Control	8	9	0	0	0
Triphasil	8	8	0	3	0
EE	10	10	1	0	0
LN	18	2	0	0	1

Treatment significantly affected the number of animals in each category (chi-squared p =
0.004). Levonorgestrel alone increased the proportion of animals assessed as "normally
cyclic".

Cycle Stage among Cycling Animals

Group	Follicular	Periovulatory	Luteal	Menstrual
Control	4	0	3	1
Triphasil	1	2	5	0
EE	5	1	1	1
LN	6	2	3	5

Treatment did not significantly alter number of animals in each cycle stage. However, the
greatest number of animals assessed as "menstrual" was in the LN-treated group; this
probably represents a withdrawal-bleeding phenomenon since animals were necropsied in
the "off week" of OCP treatment.

<u>Effects of Treatment on Caspase Expression</u>
Data tabulated below demonstrate that both glandular and stromal apoptosis was most
abundant in those animals treated with progestin-alone oral contraceptives.

Cleaved Caspase 3 in Glandular Epithelium of Endometrial Functionalis (Superficial Glands)

Group	Number	Mean Score	Std Dev	Std Err Mean	All-pairwise t tests
Ctl	19	0.58	0.90	0.21	A
Tri	19	0.63	0.60	0.14	AB
EE	21	0.24	0.54	0.12	A
LN	19	1.16*	1.12	0.26	B

ANOVA p = 0.0077
Kruskal Wallis p = 0.0136
* = different from controls at p<0.05 by Student's t test
§ rows with different letters are different by pairwise t-test

Cleaved Caspase 3 in Stroma of Endometrial Functionalis (Superficial Stroma)

Group	Number	Mean Score	Std Dev	Std Err Mean	All-pairwise t tests
Ctl	19	0.32	0.48	0.11	A
Tri	19	0.53	0.61	0.14	A
EE	21	0.33	0.58	0.13	A
LN	19	1.05*	1.08	0.25	B

Overall ANOVA p = 0.0068
Overall Kruskal Wallis p = 0.0401
* = different from controls at p<0.05 by Student's t test
§ rows with different letters are different by pairwise t-tests

TGFB3 Staining of Endometrial Functionalis

| Table of group by TGFB2 Endometrial Glands						
group	TGFB2 Immunoscore					
Frequency Row Pct	**0**	**0.5**	**1**	**2**	**3**	**Total**
Control 3 years	1 5.56	5 27.78	8 44.44	4 22.22	0 0.00	18
EE 3 years	1 4.17	6 25.00	7 29.17	10 41.67	0 0.00	24
LN 3 years	4 21.05	5 26.32	2 10.53	6 31.58	2 10.53	19 060694
Triphasil 3 years	3 15.79	4 21.05	9 47.37	2 10.53	1 5.26	19
Total	9	20	26	22	3	80
Frequency Missing = 1						

Fisher's exact test p-value is 0.1253; there is no association between TGF (B2 glands) score and treatment group.

TGFB3 Staining of Endometrial Functionalis

Table of group by TGFB2 Stroma					
group	TGFB2 Immunoscore				
Frequency Row Pct	0	0.5	1	2	Total
Control 3 years	19 100.00	0 0.00	0 0.00	0 0.00	19
EE 3 years	22 91.67	1 4.17	1 4.17	0 0.00	24
LN 3 years	15 78.95	0 0.00	3 15.79	1 5.26	19
Triphasil 3 years	18 94.74	0 0.00	0 0.00	1 5.26	19
Total	74	1	4	2	81

Fisher's exact test p-value is 0.1616; there is no association between TGF1 (B2 stroma) score and treatment group.

Figure 1: Immunostaining of TGFB3 in the macaque endometrium
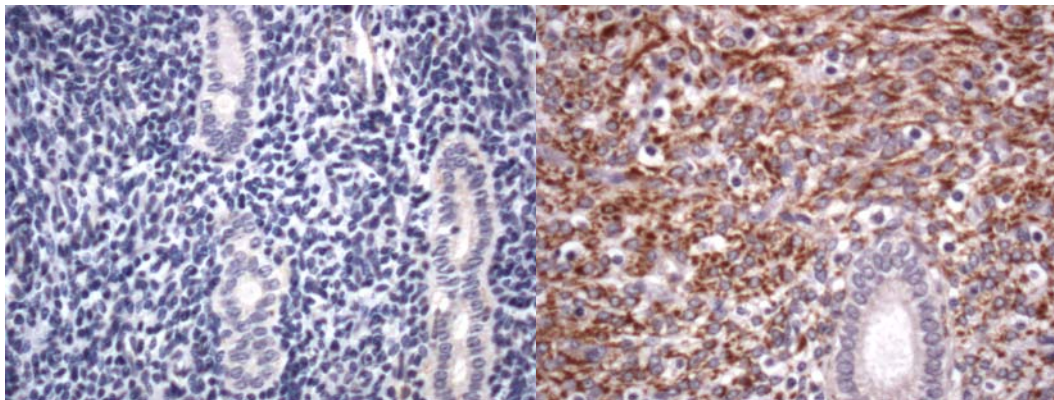
Conclusions
- Endometrial glandular and stromal apoptosis was most pronounced in animals given progestin-alone oral contraception.
- Endometrial glandular and stromal apoptosis followed the expected pattern with respect to endometrial histology, being most evident in animals with menstrual histology.
- As compared to controls, TGFBeta-1 and TGFBeta-2 and TGFBeta-3 expression was not significantly altered in the endometrial glands or stroma in premenopausal animals receiving EE or LN, administered either alone or in combination. Examples of portions of the data have been provided

Nonhuman Primate Hormone Replacement Study

Methods

Premenopausal, female cynomolgus monkeys
⬇
Oophorectomy to produce surgically-postmenopausal animals
⬇
Randomization on the basis of body weight to 4 groups:

Treatment		N
Oophorectomy + no treatment (Controls)	Group 1	25
Oophorectomy + conjugated equine estrogens (CEE) at 0.625 mg/woman/day equivalent	Group 2	25
Oophorectomy + medroxyprogesterone acetate (MPA) at 2.5 mg/woman/day equivalent	Group 3	25
Oophorectomy + CEE+MPA	Group 4	25

Treatment was given for 35 months. Animals received a moderately atherogenic diet.
⬇
Euthanasia and necropsy.
⬇
Tissues were fixed in 4% paraformaldehyde and processed into paraffin for routine histology and immunohistochemistry.

Results

Routine Histology
All slides were examined by a board-certified veterinary pathologist in consultation with MD gynecologic pathologists. Standard diagnostic terminology was used.

Group	Atrophy	Inactive	Simple Hyperplasia	Stromal Hyperplasia
Control	19	0	1	0
CEE	1	4	20	0
MPA	19	0	0	0
CEE+MPA	1	3	4	18

Endometrial Histomorphometry
Endometrial thickness and glandular area fraction were measured by computer-assisted image analysis using our published methods (Cline et al., 2001)
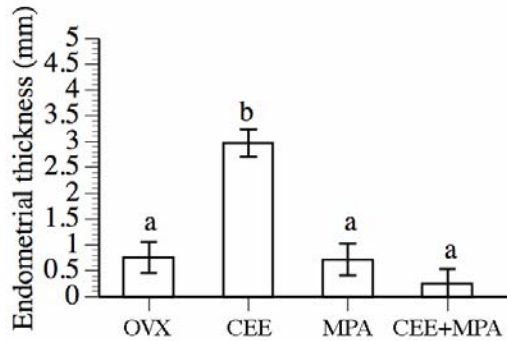
Figure 2. Endometrial thickness in millimeters (single layer thickness) in surgically-postmenopausal cynomolgus monkeys given hormone replacement therapy. OVX = control oophorectomized animals; CEE = conjugated equine estrogens; MPA= medroxyprogesterone acetate. Error bars are standard error. Bars with different letters are statistically different at p<0.05.
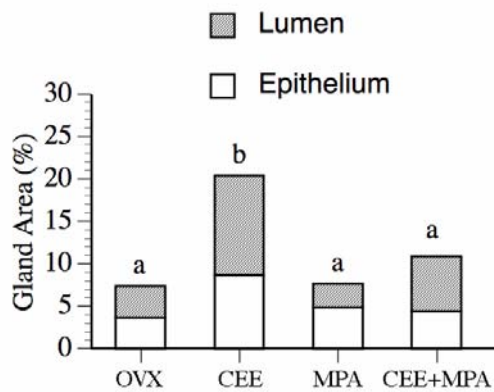

Figure 3. Endometrial glandular and luminal structures as percentages of the total endometrium in surgically-postmenopausal cynomolgus monkeys given hormone replacement therapy. OVX = control oophorectomized animals; CEE = conjugated equine estrogens; MPA= medroxyprogesterone acetate. Error bars are standard error. Bars with different letters are statistically different at p<0.05.

Analysis of Caspase Expression by Treatment Group

Immunostaining for Cleaved Caspase 3 in Superficial Glands of the Endometrium (functionalis)

Group	Number	Mean Score	Std Dev	Std Err Mean	All-pairwise t tests§
Control	22	0.09	0.43	0.09	A
CEE	24	0.58	0.72	0.15	B
MPA	18	0.39	0.61	0.14	AB
CEE+MPA	23	0.48	0.79	0.16	AB

Overall ANOVA was not significant, p = 0.0772
§ = rows with different letters are different at p<0.05 by t test

Cleaved Caspase 3 in Stroma of Endometrial Functionalis (Superficial Stroma)

Group	Number	Mean Score	Std Dev	Std Err Mean	All-pairwise t tests
Control	22	0	0	0	A
CEE	24	0.33	0.48	0.10	A
MPA	18	0.28	0.57	0.14	A
CEE+ MPA	26	1.39*	1.23	0.24	B

Overall ANOVA p < 0.0001
* = different from controls at p<0.05 by Student's t test
§ rows with different letters are different by all-pairwise t-tests

Analysis of Caspase Expression by Histologic Pattern

Immunostaining for Cleaved Caspase 3 in Superficial Glands of the Endometrium (functionalis)

Pattern	Number	Mean	Std Dev	Std Err Mean	All-pairwise t tests
Atrophic	39	0.23	0.48	0.08	A
Inactive	8	0.88	1.13	0.40	B
Simple hyperplasia	23	0.70	0.76	0.16	B
Stromal hyperplasia	15	0.13	0.35	0.09	A

Overall ANOVA p = 0.0031
§ rows with different letters are different by pairwise t-tests AT P<0.05

Cleaved Caspase 3 in Stroma of Endometrial Functionalis (Superficial Stroma)

Pattern	Number	Mean	Std Dev	Std Err Mean	All-pairwise t tests
Atrophic	39	0.15	0.43	0.07	A
Inactive	8	0.25	0.46	0.16	A
Simple hyperplasia	23	0.39	0.58	0.12	A
Stromal hyperplasia	18	1.78	1.22	0.29	B

Overall ANOVA p < 0.0001
§ rows with different letters are different by pairwise t-tests AT P<0.05

Ancillary Outcomes
Proliferation (Ki67 expression), Progesterone Receptor expression, and Estrogen
Receptor Alpha expression were measured using our standard techniques (Cline et al.,
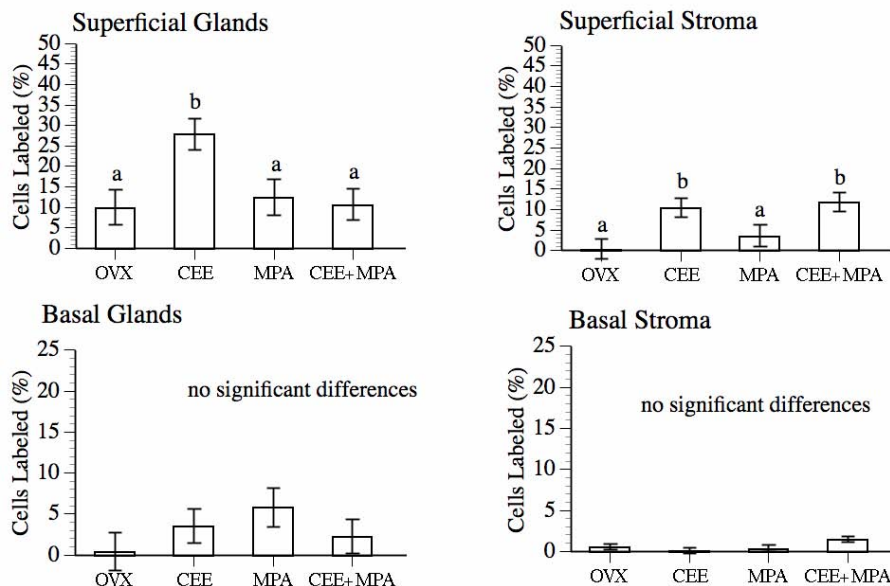2001). See next page.

Figure 4. Endometrial expression of the proliferation marker Ki67 in surgically-postmenopausal cynomolgus monkeys given hormone replacement therapy. OVX = control oophorectomized animals; CEE = conjugated equine estrogens; MPA= medroxyprogesterone acetate. Error bars are standard error. Bars with different letters are statistically different at p<0.05.
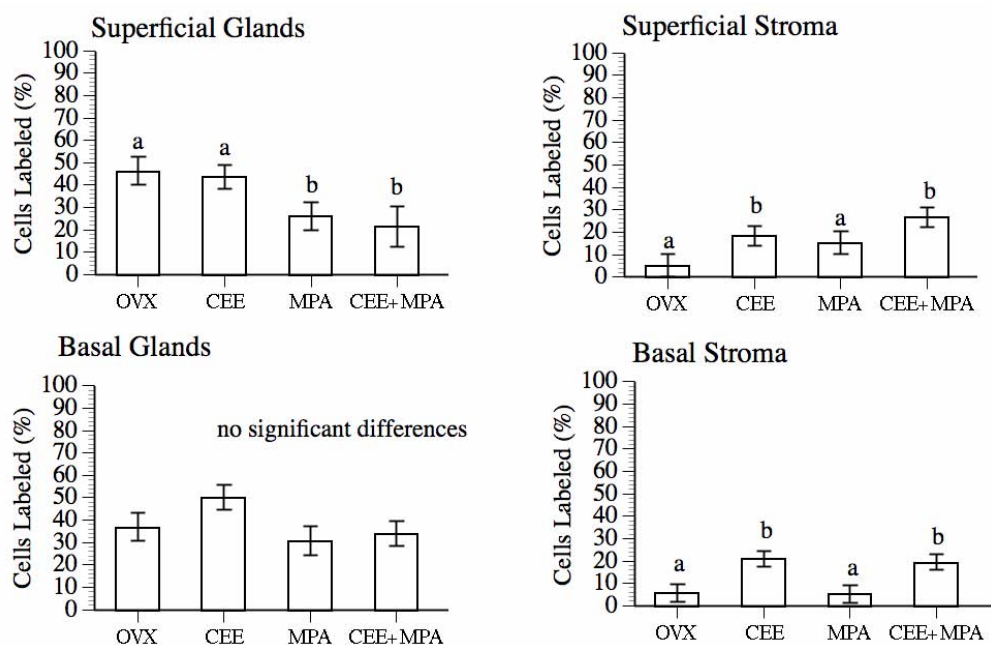

Figure 5. Endometrial expression of progesterone receptor in surgically-postmenopausal cynomolgus monkeys given hormone replacement therapy. OVX = control oophorectomized animals; CEE = conjugated equine estrogens; MPA= medroxyprogesterone acetate. Error bars are standard error. Bars with different letters are statistically different at p<0.05.
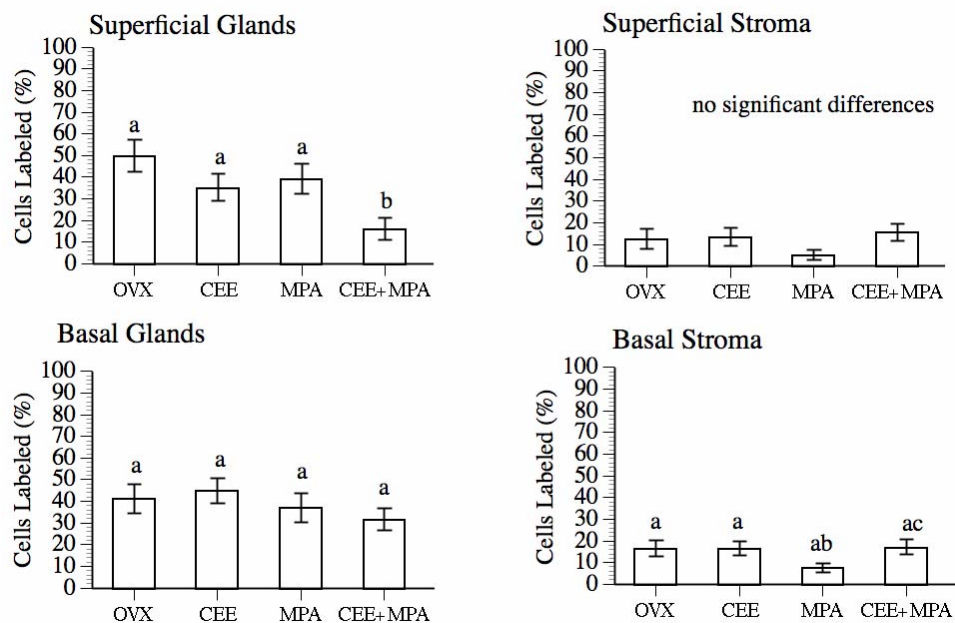
Figure 6. Endometrial expression of estrogen receptor alpha in surgically-postmenopausal cynomolgus monkeys given hormone replacement therapy. OVX = control oophorectomized animals; CEE = conjugated equine estrogens; MPA= medroxyprogesterone acetate. Error bars are standard error. Bars with different letters are statistically different at p<0.05.

TGFB3 Staining of Endometrial Functionalis

Table of group by TGFB2 Endometrial Glands						
group	TGFB2 Immunoscore					
Frequency Row Pct	**0**	**0.5**	**1**	**2**	**3**	**Total**
CEE	15 60.00	1 4.00	8 32.00	0 0.00	1 4.00	25
CEE+MPA	17 62.96	1 3.70	4 14.81	3 11.11	2 7.41	27
MPA	11 57.89	2 10.53	5 26.32	1 5.26	0 0.00	19
OVX Control	10 50.00	1 5.00	8 40.00	1 5.00	0 0.00	20
Total	53	5	25	5	3	91
Frequency Missing = 5						

Fisher's exact test p-value is 0.6356; there is no association between TGF (B2 glands) score and treatment group.

Table of group by TGFB2 Endometrial Stroma					
group	**TGFB2 Immunoscore**				
Frequency Row Pct	**0**	**0.5**	**1**	**2**	**Total**
CEE	25 100.00	0 0.00	0 0.00	0 0.00	25
CEE+MPA	11 36.67	2 6.67	9 30.00	8 26.67	30
MPA	18 94.74	0 0.00	0 0.00	1 5.26	19
OVX Control	21 100.00	0 0.00	0 0.00	0 0.00	21
Total	75	2	9	9	95
Frequency Missing = 1					

Fisher's exact test p-value is less than 0.0001; there is association between TGFB2 stromal expression and treatment; namely expression is increased significantly in the stroma in the primates receiving combined CEE+MPA therapy.

Conclusions:
- Treatment with conjugated equine estrogens (CEE) increased endometrial thickness and produced simple endometrial hyperplasia and increased endometrial glandular proliferation, as expected.
- The glandular proliferative effect of CEE was antagonized by the addition of medroxyprogesterone acetate (MPA) treatment, and induced stromal hyperplasia.
- Apoptosis was most pronounced in all endometrial compartments in animals given CEE+MPA.
- Apoptosis was significantly increased in the glands of animals with simple hyperplasia, and in the stroma of animals with stromal hyperplasia.
- In post-menopausal animals receiving CEE or MPA, administered alone or in combination. TGFBeta-2 and TGFBeta-3 expression was increased significantly in the stroma among animals receiving the combined treatment.

KEY RESEARCH ACCOMPLISHMENTS

- Aim 1: Identified genes that are differentially expressed between endometrioid and papillary serous endometrial carcinoma and determined that histology can be predicted on the basis of gene expression in approximately 90% of cases. Identified additional genes that are differentially expressed in endometrial cancer vs. normal endometria. Confirmed that microsatellite stable endometrial cancers have unique gene expression profiles compared to those with microsatellite instability
- Aim 3: Established that progestin containing oral contraceptives (OCs) are associated with a decreased endometrial cancer risk and that higher progestin- potency OCs may be more protective than lower progestin potency OCs among women with a larger body habitus.
- Aim 4: Completed analysis of apoptosis and TGF using endometrium specimens from macaques exposed to various hormonal regimens and found evidence to explain the chemoprotective effects of progestin on the pre-menopausal and post-menopausal endometrial lining

REPORTABLE OUTCOMES (since last report)

- Risinger JI, Maxwell GL, Chandramouli GV, Aprelikova O, Litzi T, Umar A, Berchuck A, Barrett JC: Gene Expression profiling of microsatellite unstable and microsatellite stable endometrial cancer indicates distinct pathways of aberrant signaling. Cancer Res. 2005 Jun; 65(12):5031-7.
- Maxwell GL**,** Chandramouli GV, Dainty L, Litzi T, Berchuck A, Barrett JC, Risinger JI: Microarray analysis of endometrial carcinomas and mixed mullerian tumors reveals distinct gene expression profiles associated with different histologic types of uterine cancer. Clin Cancer Res 2005 Jun;11(11):4056-66.
- Maxwell GL, Schildkraut JM, Calingaert B, Risinger JI, Dainty L, Marchbanks PA, Berchuck A, Barrett JC, Rodriguez GCProgestin and estrogen potency of combination oral contraceptives and endometrial cancer risk. Gynecol Oncol. 2006 May 30; [Epub ahead of print]

CONCLUSIONS

Different histological types of cancer have genomic expression patterns that reflect unique pathways of carcinogenesis. Likewise, cancers characterized by microsatellite instability result in the expression of genes most likely to be affected by alterations in mismatch repair. As we improve our understanding of the alterations that accompany endometrial carcinogenesis, it is likely that future chemopreventives may be developed for several types of endometrial cancer, each of which develops by specific pathways. In regards to contemporary chemopreventive options, an analysis of data from the CDC CASH database has suggested that a greater protective effect against endometrial cancer may be associated with high progestin potency OCs, particularly in patients with a larger body habitus. Higher potency progestin containing OCs should be considered in forthcoming endometrial cancer prevention trials particularly if other studies suggest a greater risk reduction associated with heavier women that are at highest risk for endometrial cancer. Using a macaque model, we have determined that the mechanisms behind the chemoprotective effects of progestin containing hormonal regimens in both pre-menopausal and post-menopausal patients appear to be in part related to induction of apoptosis. We look forward to continued ongoing interpretation of our data as well as evaluation of the short term effects of progestin

containing hormonal formulations in our clinical trial evaluating the short term effects of progestin on the endometrium lining using both a targeted analysis of apoptosis as well as an assessment of global gene expression using oligonucleotide microarray.

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